

Applicants: Desiree H.H. Tsao, Jean-Baptiste Telliez,
Thomas McDonagh, Lih-Ling Lin, Sang Hsu,
Guang-Yi Xu, and A. Karl Malakian
Serial No.: 09/821,819
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Please replace the paragraph at page 30, line 8 with the following:

A2
The CD40 peptide (SNTAAPVQETLHG-OH) (SEQ ID NO:2) was synthesized by using fluorenylmethoxycarbonyl (Fmoc) solid-phase methods and purified by reverse-phase HPLC.

Please replace the paragraph at page 34, line 14 with the following:

A3
Inhibition of N-TRADD/C-TRAF2 by CD40-derived peptide: Recent reports on a C-TRAF2 binding peptide derived from CD40 receptor (Pullen, *et al.*, Biochemistry 37: 11836-11845, 1998; Sato, *et al.*, FEBS Lett 358: 113-118, 1995; Nakano, *et al.*, J. Biol. Chem. 271: 14661-14664, 1996) led the inventors to study the effect of this peptide on the N-TRADD/C-TRAF2 interaction. CD40 belongs to the TNF receptor family and has been shown to interact with several TRAF family members by yeast two hybrid analysis and co-precipitation assays (Pullen, *et al.*, J. Biol. Chem. 274: 14246-14254, 1999; Pullen, *et al.*, Biochemistry 37: 11836-11845, 1998; Cheng, *et al.*, Science 267: 1494-1498, 1995). In particular, full length TRAF2 has been shown to interact directly with the CD40 cytoplasmic domain (Pullen, *et al.*, Biochemistry 37: 11836-11845, 1998). The binding site of CD40 for TRAF2 was defined by peptide mapping where the shortest CD40 sequence that TRAF2 recognized was a five amino acid peptide with the sequence PVQET (amino acid residues 6-10 of SEQ ID NO:2). The crystal structure of C-TRAF2 (311-501) with a peptide derived from CD40 with sequence YPIQET (SEQ ID NO:3) (designated CD40-p1) (McWhirter, *et al.*, Proc. Natl. Acad. Sci. USA 96: 8408-8413, 1999) (Published with Protein Data Bank at Accession No. 1QSC, and expressly incorporated herein by reference) shows that it binds each of the TRAF2 monomers in the C-TRAF2 trimer complex. Comparison with the structure of the peptide from TNFR-2 in complex with C-

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A3
CD40
TRAF2 (Park, *et al.*, Nature 398: 533-538, 1999), which has a different consensus sequence (QVPFSKEEC) (SEQ ID NO:4), reveals similar affinities and conformations (McWhirter, *et al.*, Proc. Natl. Acad. Sci. USA 96: 8408-8413, 1999). However, despite similar backbone contacts, the two peptides are slightly shifted in the binding site, where CD40 peptide makes many more complementary contacts with C-TRAF2 than does the TNFR-2-derived peptide.

Please replace the paragraph at page 35, line 12 with the following:

A4
In the instant studies, the interaction between a 13-mer peptide derived from CD40 (a longer version of CD40-p1, with sequence SNTAAPVQETLHG (SEQ ID NO:2)) with C-TRAF2 was characterized, as well as its effect on the N-TRADD/C-TRAF2 interaction. BIAcore studies show that the peptide binds to C-TRAF2 with an affinity of ~1.0 mM (data not shown). In addition the peptide was also able to compete for the binding of N-TRADD to C-TRAF2, with an IC50 of ~ 1mM.

Please replace the paragraph at page 36, line 27 with the following:

A5
Based on previous studies (Arch, *et al.*, Genes Dev 12: 2821-2830, 1998; Park, *et al.*, Nature 398: 533-538, 1999; McWhirter, *et al.*, Proc. Natl. Acad. Sci. USA 96: 8408-8413, 1999), C-TRAF2 can recognize at least two different sequence motifs, SXXE (SEQ ID NO:5) in the case of TNFR2 and PXQXT (SEQ ID NO:6) for CD40. Although the two peptides make similar backbone contacts, each peptide makes different additional unique contacts with C-TRAF2, suggesting the presence of distinct recognition sites. Neither consensus sequence is present in N-TRADD, implying that there may be another